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EXAMINER	
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ART UNIT	PAPER NUMBER
1635	

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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/505,482	STREBHARDT ET AL.	
	Examiner	Art Unit	
	Louis V. Wollenberger	1635	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *22 May 2006 and 25 August 2006*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *32-64* is/are pending in the application.

4a) Of the above claim(s) *57-62 and 64* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *32-56 and 63* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Sequence Listing/Compliance

Applicant's submission of a substitute sequence listing in the reply filed 8/25/06 in paper and computer readable form is acknowledged.

The CRF has been entered into the application.

Also acknowledged are the amendments to the specification submitted with the reply filed 5/22/06.

With these submissions, the application is now considered to be in sequence compliance.

Election/Restrictions

Applicant's election with traverse of Group I, claim(s) 32–56 and 63, drawn to an RNA for inhibiting development or progress of proliferative diseases, wherein said RNA reduces or inhibits the activity of mammalian polo like kinase (PLK 1), and to a phosphorothioate antisense oligonucleotide (ASO), and to the ASO corresponding to **SEQ ID NO:30**, as recited in claim 56, in the reply filed on 8/25/06 is acknowledged.

The traversal is on the ground(s) that the agent of claim 32 was already searched by the Office as part of the Non-Final Office Action of 12/20/05. Applicants further submit that Groups I and II share special technical features in that they are both agents for inhibiting the development or progress of proliferative diseases, cancer diseases or other diseases which are accompanied by elevated PLK1 expression levels, they both reduce or inhibit the activity of mammalian polo like kinase 1 in mammalian cells and they both include at least one

phosphorothioate antisense oligonucleotide.

This is not found persuasive because although the content of claim 32 was searched in the previous Office Action, neither SEQ ID NO:30 nor 31 were searched in the previous Office Action. Furthermore, burden of search is irrelevant to Restriction practice in a National Stage Application submitted under 35 USC 371(c). Restriction in this case is based on the fact that, although Groups I and II share common features, but they do not share a special or corresponding technical feature.

Pursuant to PCT Rule 13.2—Circumstances in Which the Requirement of Unity of Invention Is to Be Considered Fulfilled—Where a group of inventions is claimed in one and the same international application, the requirement of unity of invention referred to in Rule 13.1 shall be fulfilled only when there is a technical relationship among those inventions involving one or more of the same or corresponding special technical features. The expression "special technical features" shall mean those technical features that define a contribution which each of the claimed inventions, considered as a whole, makes over the prior art.

In the instant case, agents, including antisense nucleic acids, that inhibit or reduce the activity of PLK1 in mammalian cells" were known in the art at the time the instant invention was made, as evidenced by the prior art cited in the Action of 12/20/05 and in the original Restriction Requirement of 7/25/06 (Wolf et al.). Accordingly, agents that inhibit PLK1 cannot be a special technical feature.

As explained in the foregoing restriction requirement of 7/26/06, the special technical feature of Group I is a phosphorothioate antisense oligonucleotide (ASO) corresponding to SEQ ID NO:30, whereas the special technical feature of Group II is a phosphorothioate antisense

oligonucleotide (ASO) corresponding to SEQ ID NO:31. Accordingly, unity of invention is lacking *a priori.*

The requirement is still deemed proper and is therefore made FINAL.

Status of Application/Amendment/Claims

Applicant's response, filed 5/22/2006, to the Non-Final Office Action, mailed on 12/20/05, is acknowledged.

Also acknowledged are Applicants' amendments to the claims.

With entry of the amendment filed on 8/25/06, Claims 32–64 are pending.

Claims 57–62 and 64 are withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to a nonelected invention, there being no allowable generic or linking claim. Applicant timely traversed the restriction (election) requirement in the replies filed on 10/24/05 and 8/25/06.

Claims 32–56 and 63 are currently under examination. Applicants have expressly elected "siRNA4" (claim 36) and SEQ ID NO:30 (claim 56).

This application contains claims drawn to an invention nonelected with traverse; specifically, claims 57-62; SEQ ID NO:31; and siRNAs 2, 3, and 5. A complete reply to the final rejection must include cancellation of nonelected claims or other appropriate action (37 CFR 1.144) See MPEP § 821.01.

Claim Objections

Claims 33-36 and 53-56 are objected to for the following reasons.

Claims 33-36 and 53-56 recite "as active agent" in the final lines of claims 33 and 53. It is not entirely clear whether "active agent" is referring to "the PLK1 gene" or the inhibitory RNA. The term "active agent" lacks clear antecedent basis. Claims 34-36 and 54-56 are objected to on that basis because of their dependence on claim 33 and 53, respectively. Clarification and/or correction is required.

Claims 36 and 56 are further objected to for reciting non-elected inventions. Specifically, claim 36 recites siRNAs 2, 3, and 5. Claim 56 recites SEQ ID NO:31. No generic claim is currently allowable. A complete reply to the final rejection must include cancellation of these nonelected inventions or other appropriate action (37 CFR 1.144) See MPEP § 821.01.

Claim Rejections - 35 USC § 112

Claims 36, 40, 51, and 53-56 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 36 recites a series of siRNA molecules, wherein the siRNA sequences are specified according to the complementary positions in "the PLK1 gene." The claim is indefinite because the sequences of the claimed siRNA, siRNA4 (elected with traverse) cannot be determined from the instant claim—i.e., there is no reference point or context for the recited "positions" because the PLK1 gene sequence is not set forth in the claim. Accordingly, the Examiner is unable to interpret the claim with certainty (MPEP §2173.06). Accordingly, no art rejection can be made on claim 36 as currently written. The instantly claimed siRNA is more properly recited with the use of a SEQ ID NO: identifier.

Claim 40 recites the phrase "short antisense RNA." The term "short" is a relative term which renders the claim indefinite. The term "short" as applied to antisense is not defined by the claim, the specification does not provide a standard for ascertaining the requisite degree, and one of ordinary skill in the art would not be reasonably apprised of the scope of the invention. While the term "short interfering RNA" or "siRNA" may be considered to be a term of art in view of the disclosure of Elbashir et al. (2001) *Nature* 411:494-498, the term "short antisense RNA" is not a recognizable term of art. Antisense RNAs may be of almost any length. Accordingly, the metes and bounds of the claim are unclear.

Clarification and/or correction is required.

Claim 51 recites the limitation "the active substances." There is insufficient antecedent basis for this limitation in the claim.

Claim 53 recites "The agent according to claim 32, wherein said agent contains at least one phosphorothiate antisense oligonucleotide (ASO) or an ASO with another modification like mixed backbone oligonucleotides or morpholino oligonucleotides directed against the PLK 1 gene as active agent."

The phrase "like mixed backbone oligonucleotides or morpholino oligonucleotides" renders the claim(s) indefinite because the claim includes elements not actually disclosed (those encompassed by "like"), thereby rendering the scope of the claim(s) unascertainable. See MPEP § 2173.05(d). That is, the modifications may be interpreted to be either "mixed backbone" or "morpholino" or "like" "mixed backbone" or "morpholino." However, neither the claim nor the specification describes what modifications are "like" mixed backbone" or "morpholino." Claims 54-56 are rejected on this basis due to their dependence on claim 53.

Appropriate correction is required.

***

Claims 32-35, 37-55, and 63 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. This is a written description rejection.

Factors to be considered in determining whether there is sufficient evidence of possession include the level of skill and knowledge in the art, complete or partial structure, physical and/or chemical properties, functional characteristics alone or coupled with a known or disclosed correlation between structure and function, and the method of making the claimed invention. Disclosure of any combination of such identifying characteristics that distinguish the claimed invention from other materials and would lead one of skill in the art to the conclusion that the applicant was in possession of the claimed species is sufficient.

With the amendment of 8/25/06, the claims are drawn to RNA and antisense agents that inhibit or reduce the activity of mammalian polo like kinase 1 (PLK1) in mammalian cells ant that thereby inhibit the development or progress of proliferative diseases such as cancer.

The claims are broad in that they encompass any RNA molecule, including any ribozyme, antisense nucleic acid, hairpin RNA, or siRNA, and any DNA antisense oligonucleotide or triplex forming oligonucleotide, for example, having the ability to target and inhibit any form of mammalian PLK1, including any variant or isoform, from

any mammalian organism. Furthermore, the claims require not only that the RNA and antisense agents inhibit PLK1 activity, but that they also have a specific biological effect, as now recited, for inhibiting the development or progress of proliferative diseases, cancer diseases, or of other diseases which are accompanied by elevated PLK1 expression levels.

It is important to note that the claimed RNA and DNA agents act not simply as probes or primers but as effectors of biological activity in a cell. The agents must not only hybridize or share substantial complementarity to the intended PLK1 target but also produce the desired inhibitory effect in the cell in a mammal, including reduction of gene expression and inhibition of a proliferative disease. Accordingly, there is a structure-function relationship that is critical to the claimed invention.

With regard to siRNA agents, embraced by the instant claims, post-filing art teaches that complementarity alone does not guarantee RNAi activity in a cell; other factors, including sequence accessibility and base composition, appear to influence the activity of an siRNA. These factors were not well understood at the time the instant application was filed.

For instance, Holen et al. (2002) *Nucleic Acids Res.* 30:1757–1766 tested several siRNAs corresponding to different target sequences in human coagulation trigger tissue factor (hTF) for their ability to induce silencing of the hTF gene. Of the several siRNAs synthesized and tested only a few produced significant reduction in expression of hTF, suggesting that accessible siRNA target sites may be rare in some human mRNAs. Moreover, siRNAs targeting different

sites in hTF demonstrated dramatic differences in silencing potency. Although strong positional effects were observed and regions of high GC content seem to be targeted less efficiently than those of low GC content, Holen et al. concluded that the factors determining the differences in siRNA efficiency remain unclear and that susceptible RNAi target sites in some human genes may be rare.

The results of Holen et al. suggest that it is difficult to predict *a priori* what sequences to target in a gene with siRNAs to induce efficient silencing by RNAi. In addition, there is growing body of evidence suggesting that specific siRNAs may produce unwanted and unanticipated off-target effects (see Jackson et al. 2003, *Nature Biotechnology* 21:635-637). Their results indicate that treating cells with different siRNAs targeting different sequences in the same RNA transcript may result in different but reproducible off-target effects.

Accordingly, the art indicates that researchers must empirically determine which sequences and their corresponding siRNAs provide for the intended biological and therapeutic effects.

Thus, in view of these teachings, it can be concluded that variability in Applicants' instantly claimed genus clearly exists. In view of the art-recognized variability in siRNA function and efficacy, one of skill in the art would need to look to the instant priority document, to which benefit is claimed, for a description of the siRNAs that inhibit mammalian PLK1 genes.

MPEP §2163 states in part that

> "The written description requirement for a claimed genus may be satisfied through sufficient description of a representative number of species by actual reduction to practice (see i)(A), above), reduction to drawings (see i)(B), above), or by disclosure of relevant, identifying characteristics, i.e., structure or other physical and/or chemical properties, by functional characteristics coupled with a known or disclosed correlation between function and structure, or by a

> combination of such identifying characteristics, sufficient to show the applicant was in possession of the claimed genus (see i)(C), above). See *Eli Lilly*, 119 F.3d at 1568, 43 USPQ2d at 1406."

The instant application does not satisfy either of these criteria with regard to the claimed RNA and antisense reagents in that the application does not describe a representative number of species having the intended inhibitory and biological functions nor a feature common to all agents in the genus that inhibit any mammalian PLK1 and thereby inhibit the development or progress of proliferative diseases, cancer diseases, or of other diseases which are accompanied by elevated PLK1 expression levels.

While the claims are directed to agents targeting only mammalian PLK1, the structural identity of these targets have not been clearly set forth, aside from that sequence corresponding to human PLK1 (see page 33 of specification, for example). Further, it is unclear from the prior art whether one of skill would be reasonably apprised of the sequences of the complete genus of target genes comprising the mammalian PLK1 family, which may be both structurally and functionally diverse when compared among the many thousands of possible mammalian species in existence.

Thus, because the instant claims require a specific biological and therapeutic function for each of the many thousands of different RNA and DNA agents now embraced by the claims, and because applicant has described neither a sufficient number of species representative of the genus nor a feature common to the genus, it is the Examiner's position that adequate written description does not exist in the instant application for all these RNA and DNA nucleic acid agents for inhibiting any mammalian PLK1 and inhibiting any proliferative disease.

Vas-Cath Inc. v. Mahurkar, 19USPQ2d 1111, clearly states that applicant must convey with reasonable clarity to those skilled in the art that, as of the filing date sought, he or she was in possession of the invention. The invention is, for purposes of the written description inquiry, whatever is now claimed (pg. 1117).

An applicant shows possession of the claimed invention by describing the claimed invention with all of its limitations using such descriptive means as words, structures, figures, diagrams, and formulas that fully set forth the claimed invention. *Lockwood v. American Airlines, Inc.*, 107 F.3d 1565, 1572, 41 USPQ2d 1961, 1966 (Fed. Cir. 1997).

While the specification adequately describes certain specific inhibitory RNAs such as siRNAs and antisense oligonucleotides (see in particular pages 33 and 66 and Table 1) directed against the human PLK1 gene corresponding to GenBank Accession No. X75932, by fully setting forth their sequences and describing their functions, and by describing the materials and methods needed to make and use these agents, adequate written description does not exist for the virtually unlimited number of other siRNAs, antisense oligos, and ribozymes against all other mammalian PLK1 genes.

Applicants have not shown possession of the entire genus. Rather applicants describe a narrow subset of agents; specifically, certain structurally defined siRNAs and antisense oligos against human PLK1, GenBank Accession No. X75932.

MPEP §2163 states, in part: "[A] patentee of a biotechnological invention cannot necessarily claim a genus after only describing a limited number of species because there may be unpredictability in the results obtained from species other than those specifically enumerated. A patentee will not be deemed to have invented species sufficient to constitute the genus by virtue

of having disclosed a single species when ... the evidence indicates ordinary artisans could not predict the operability in the invention of any species other than the one disclosed. *In re Curtis*, 354 F.3d 1347, 1358, 69 USPQ2d 1274, 1282 (Fed. Cir. 2004)."

Accordingly, only methods comprising the use of the disclosed structural and functionally defined, human PLK1 gene-specific inhibitory siRNAs, shRNAs, and antisense oligonucleotides meet the written description requirement.

Applicant is reminded that the written description requirement is separate and distinct from the enablement requirement. *In re Barker*, 559 F.2d 588, 194 USPQ 470 (CCPA 1977), cert. denied, 434 U.S. 1064 (1978); *Vas-Cath, Inc.* v. *Mahurkar*, 935 F.2d 1555, 1562, 19 USPQ2d 1111, 1115 (Fed. Cir. 1991).

Applicants' arguments:

Arguing the rejection of previous claims 1-23 and 30 (remarks 5/22/06), Applicants state that numerous examples of RNA agents are described at pages 33-78 of the application and that one of skill in the art would reasonably expect any and all RNA agents that inhibit polo like kinase activity in mammalian cells to work in the present invention.

Applicants' remarks have been fully considered but are not found persuasive.

While one of skill may reasonably expect certain classes of molecules to work in any given embodiment now encompassed by the instant claims, that fact alone does not help to describe the partial or complete structure of the claimed agents themselves.

A description which renders obvious a claimed invention is not sufficient to satisfy the written description requirement of that invention. *Regents of the Univ. of Cal. V Eli Lilly & Co., Inc.* 43 USPQ2d 1398, citing *Lockwood* , 107 F.3d at 1572, 41 USPQ2d at 1966.

Accordingly, the instant claims are rejected for lack of written description support.

Claim Rejections - 35 USC § 102

Claims 32–35, 53-55, and 63 are rejected under 35 U.S.C. 102(b) as being anticipated by Elez et al. (2000) *Biochemical and Biophysical Res. Comm.* 269:352–356.

Applicants are advised that as amended claim 32 is considered to encompass both RNA and DNA antisense agents targeting mammalian PLK1. Broadest reasonable interpretation of the recitation "antisense oligonucleotide" includes antisense oligonucleotides comprising RNA and/or DNA nucleotides, as well as chemically modified variants thereof.

Furthermore, claim 33 does not clearly limit the invention of claim 32 to RNA agents, since it recites only that the "agent according to claim 32" contains at least one RNA. The term "contains" is considered to be open language, which does not clearly preclude the presence of other inhibitory agents such as the antisense oligonucleotide recited in claim 32.

Elez et al. teach a 19-mer antisense phophorothioate-modified oligonucleotide, JWG2000, that inhibits PLK1 expression in culture in human carcinoma cell lines A549 and Detroit 562, and *in vivo* in A549 xenografts in nude mice (pp. 352–355, Figs. 1, 2, and 4). The specific sequence of the exemplary antisense oligo, JWG2000, is disclosed on page 353, second column. Upon transfection into cultured cells, the oligo is shown to reduce PLK1 mRNA and protein expression levels (Figs 1 and 2). Upon bolus delivery to nude mice, the antisense oligo is shown to have an antineoplastic effect in that tumor size is decreased. The authors state (page 354) that down-modulation of PLK1 expression with JWG2000 was accompanied with decreased proliferation and viability of cancer cells, and a decrease in tumor size in treated

animals. For administration to mice, the oligonucleotide was dissolved in saline solution (page 353) to produce a pharmaceutical composition within the scope of claim 30.

Accordingly, the instant claims are anticipated by Elez et al.

Claim Rejections - 35 USC § 103

Claims 32–35, 37, 39-44, 46, 49-51, 53-55, and 63 are rejected under 35 U.S.C. 103(a) as being unpatentable over Holtrich et al. (1994); Elez et al. (2000); and Driscoll et al. (WO 01/49844 A1).

Holtrich et al. (1994) teach the cDNA sequence of human polo-like kinase, deposited as GenBank accession no. X75932. Thus, the human *plk* sequence is shown in the prior art. Holtrich et al. do not teach an antisense RNA, siRNA, or hairpin RNA expression construct for inhibiting the expression of human PLK1.

Elez et al. is relied upon for the reasons given above. Elez et al. teach an antisense sequence for down regulating the *plk1* gene, thereby reducing PLK1 activity. Elez et al. further teach that Plk1 is a highly conserved mitotic serine/threonine kinase that is overexpressed in cancer cell lines and that PLK1 could serve as a suitable diagnostic and prognostic marker for tumor progression and as target for anti-cancer therapy (page 352). Moreover, interference with PLK1 expression at the mRNA level leads to loss of cell viability, blockage of tumor cell proliferation, and induction of mitotic death in Plk1-overexpressing cell lines. Thus, antisense ODNs against Plk1 mRNA may provide a novel therapeutical concept to inhibit the production of Plk1 protein via antisense-induced degradation of its mRNA. Elez et al. state that Plk1 seems an excellent target for antisense anti-tumor therapy since it plays a major role in G2/M transition

and, via Cdc25C activation and cyclin B degradation in telophase, also controls the cells' exit from mitosis (page 355).

Elez et al. further teach that antisense oligodeoxynucleotides (ODNs) offer potential not only for investigation of gene expression, but also as therapeutic agents by altering the intermediary metabolism of RNA, thus modulating transfer of information from gene to protein (page 352, 2nd column). Further, it is stated that the present study indicates that JWG2000 is a potent and specific 20-mer phosphorothioate ODN antisense inhibitor of Plk1 expression as its anti-proliferative and antitumor activity in cell culture (A549 and Detroit562) and in mouse models xenografted with A549 cells could be clearly established (page 352, 2nd column).

Driscoll et al. teach a DNA construct encoding an inverted repeat (hairpin) RNA, which is capable of binding to an mRNA sequence of interest and mediating RNA interference in vitro or in vivo (pages 1-5; Figs. 1 and 2). It is taught that the inverted repeat RNAi expression construct exploits the ability of a vector to generate multiple dsRNA copies, obviating the need for continuous administration of naked dsRNA duplexes, and providing for prolonged expression of the inhibitory RNA molecules indefinitely *in vivo*. Another advantage of the IR (inverted repeat) constructs is said to be their heritable nature, allowing for the production of transgenic animals and long term, and possibly inducible, silencing of genes (pages 8 and 25). The IR constructs are taught as having utility for the treatment of neoplastic diseases. According to Driscoll et al., the aberrant expression of oncogenes in certain cancers may be targeted for gene silencing using the compositions and methods of the invention (page 9).

Importantly, Driscoll et al. teach that double-stranded RNA is at least an order of magnitude more potent at inducing RNA interference than are preparations of either strand alone

(page 1). According to Driscoll et al., the surprising properties of dsRNA-mediated interference prompted users to abandon the term "antisense" and to begin referring to the process as "RNA interference." One of skill in the art may infer from these teaching that dsRNA, such as hairpin RNA, is more potent than antisense or sense strands alone. One of skill in the art would therefore be motivated to make and use the dsRNA expression vectors taught by Driscoll et al. to down regulate a gene of interest since the inverted repeat expression constructs and the transcripts they encode are said to have specific advantages for the down regulation of target genes.

Driscoll et al. teach that the IR expression constructs comprise a promoter element operably linked to a first coding sequence in a sense orientation, which is in turn linked to second coding sequence in an antisense orientation (page 11). The first and second coding sequences may range between 20 and 2500 nucleotides in length (page 11) and may be separated by a spacer sequence of between 300 and 1500 nucleotides in length (page 3). The promoters may be any of those listed on pages 3 and 12-13. The IR expression construct may also comprise a 3' terminator sequence (page 14). Expression of the IR gene from the promoter results in the formation of a double stranded "snap back" RNA, capable of abrogating the expression of an endogenous gene (page 25 and see Figs.1 and 2). Exemplary vectors are described in Table II and in examples I–III, pages 35–50, and shown in Figs. 4–6.

Additonally, Driscoll et al. teach that IR expression vectors are generally administered to a patient as a pharmaceutical composition (page 30). Accordingly, the pharmaceutical composition may be formulated for administration via direct injection into the brain or intravenous injection (pages 30-31). For direct injection (i.e., bolus injection) into the brain, it is taught that the vector should be dispersed in a medium similar in composition to cerebrospinal

fluid (page 31). Alternatively, the vector may be formulated in buffered saline (page 30).

It would have been obvious to one of ordinary skill in the art to use the cDNA sequence disclosed by Holtrich et al. and the baseline antisense studies of Elez et al. to generate short hairpin RNA expression vectors, as taught by Driscoll *et al.,* for inhibition of PLK1 gene expression *in vitro* and *in vivo*.

One would have been motivated to create such compounds because Elez et al. expressly teach that PLK1 could serve as target for anti-cancer therapy antisense compounds and that interfering with PLK1 expression using an antisense oligonucleotide blocks tumor cell proliferation; and because Driscoll et al. teach that dsRNA, such as hairpin RNA generated by an expression construct may be a more potent inhibitor of mRNA expression than either sense or antisense oligos alone.

One would have a reasonable expectation of success given that Driscoll et al. fully describe the materials and methods necessary to generate and use inverted repeat constructs to virtually any known gene such as polo-like kinase, whose sequence is disclosed in the prior art by Holtrich et al. Futhermore, Elez et al. teach that antisense treatment of proliferating cells expressing PLK1 is an effective therapy for reducing tumor size. Given the relative higher potency of dsRNA, as taught by Driscoll et al., one of skill would have been motivated and had a reasonable expectation of success to build and use dsRNA expressing vectors as an additional tool to study and manipulate PLK1 expression in cultured cells and live animals.

Thus in the absence of evidence to the contrary, the invention as a whole would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made.

Applicants' arguments:

Applicants arguments are specifically directed to previous claims 6-11, 13, 14, and 16-18, now cancelled. However, applicants arguments as may be related to the current claims are addessed herein to expedite prosecution.

Applicants arguments that Holtrich and Elez do not teach that an RNA, inhibitory peptide or antibody agent which reduces or inhibits the activity of polo like kinase in mammalian cells can be used to inhibit the development or progress of proliferative diseases is moot in light of Applicants' amendment to claim 32, adding the recitation "antisense oligonucleotide." This term encompasses both DNA and RNA agents.

Applicants further argue that one skilled in the art would not use Driscoll's repeat gene construct to reduce or inhibit the activity of PLKI. Applicants assert that the CMV promoter used by Driscoll leads to the transcription of sequences in addition to those needed for the shRNA. Such by products are clearly unwanted and lead to contaminated RNAs. Applicants point out that Driscoll discloses a spacer region between 300-1000 nucleotides in length, and that the longer a spacer is, the more the hairpin structures are distorted and the hybrid is consequently less stable and not as effective. Applicants contend that one skilled in the art would not be motivated to combine Holtrich and Elez with Driscoll, to reduce or inhibit the activity of PLKI because of the unspecific by products which result from Driscoll's process. In view of the above discussion, applicants request that this rejection be withdrawn.

Response to Arguments

Applicant's arguments filed 5/22/06 have been fully considered but they are not persuasive.

Applicants appear to be arguing that the materials and methods, disclosed by Driscoll et al., for inhibiting gene expression using hairpin RNA expression constructs, are inoperative, or would render the prior art combination unsuitable for its intended purpose.

The test for obviousness is what the combined teachings of the references would have suggested to one of ordinary skill in the art, and all teachings in the prior art must be considered to the extent that they are in analogous arts (MPEP §2143.01, II).

Obviousness does not require absolute predictability, however, at least some degree of predictability is required. Evidence showing there was no reasonable expectation of success may support a conclusion of nonobviousness (MPEP §2143.02).

Whether an art is predictable or whether the proposed modification or combination of the prior art has a reasonable expectation of success is determined at the time the invention was made (MPEP §2143.02).

In the instant case, Driscoll et al. provide an explicit motivation to use hairpin-expression constructs to inhibit gene expression in worms, plants, and mammals for a variety of investigational and therapeutic purposes.

Elez et al. provide ample motivation to use antisense oligonucleotides to inhibit human PLK1 in cells *in vitro* and *in vivo* to understand its function and to possibly develop suitable treatment therapies for human subjects.

Applicants' claims that the Driscoll et al. strategy to inhbit gene expression *in vivo* is unsuitable for methods in mammals is not sufficient to defeat Driscoll et al. as valid prior art teaching and suggesting the use of such RNA expression constructs at the time the instant invention was made.

Driscoll et al. provide at least one working example, albeit in worms, wherein hairpin RNAs expressed in vivo in transgenic worms according to their invention are shown to result gene expression inhibition in vivo in living worms (see Example I, beginning at page 35).

This example along with the general and detailed guidance of the specification would reasonably lead one to believe in the operability of the Driscoll et al. invention and would be sufficient to motivate one of skill in the art to make and use the invention for use in any organism including mammals, as expressly suggested by Driscoll et al.

While it may be true that Driscoll et al. point the reader towards the CMV promoter, Driscoll et al. teach that other promoters are known to those in the art and inlcude the Mt promoter, SV40 promoter, and glucocorticoid promoter (page 12).

Furthermore, Driscoll et al. expressly teach that the duplex or stem region of the hairpin may be about 20 to 2500 nucleotides in length (page 11), which encompasses the range specified in claim 43, for example.

Applicants have not provided convincing evidence that the Driscoll et al. invention, despite the recommended spacer size and/or promoter would be inoperable or absolutely unsuitable for every embodiment now encompassed by the instant claims, or that the methods of Driscoll et al. would not provide for a "reasonable" expectation of success.

Accordingly, the instant claims are rejected as being obvious over Holtrich et al. (1994); Elez et al. (2000); and Driscoll et al. (WO 01/49844 A1).

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Claim 45 is rejected under 35 U.S.C. 103(a) as being unpatentable over Holtrich et al. (1994); Elez et al. (2000); and Driscoll et al. as applied to claims 32–35, 37, 39-44, 46, 49-51,

53-55, and 63 above, and further in view of Kennerdell et al. (2000) *Nature Biotechnology* 17:896–898; and Martinek et al. (2000) *Genetics* 156:1717–1725.

Claim 45 recites an RNA expression system according to claim 37 having a spacer sequence of 3 to 10 nucleotides.

Holtrich et al. (1994); Elez et al. (2000); and Driscoll et al. are relied upon for the reasons given above. These references do not teach a IR, hairpin expression construct, wherein the complementary sequences are separated by a spacer sequence of 3 to 10 nucleotides.

Driscoll et al. teach that the inverted repeat gene may comprise a spacer, or linker sequence of between 300 and 1500 nucleotides in length (page 3).

Furthermore, the prior art is replete with teachings of hairpin expression constructs encoding stem-loop RNA (e.g., ribozymes, tRNA, and small RNAs) having various loop and stem sizes.

For example, Kennerdell et al. disclose an RNA expression construct for expressing interfering, hairpin-loop RNA targeted to a *lacZ* transgene in *Drosophila* (page 896-7, Fig. 2) The construct is said to comprise an inverted repeat gene sequence having dyad symmetry centered about a five (5)-base pair linker, expressed from an inducible expression vector (Fig. 1 and 2, legends).

Similarly, Martinek et al. disclose an inverted repeat expression construct for inhibiting endogenous gene expression in *Drosophila*, comprising a 67-nt spacer region (page 1719–20, Fig. 1).

Taken together, the teachings of Driscoll et al., Kennerdell et al., Martinek et al. imply that the loop size, or spacer sequence, separating complementary sense and antisense sequences

in an RNA expression construct is a parameter that may vary from construct to construct. Each of the groups teaches effective gene silencing using shRNAs with different loop sizes ranging from 5 to 1500 nucleotides. Kennerdell et al., in particular, teaches that the spacer sequence may be as small as five nucleotides while still providing for the formation of a hairpin RNA and effective gene silencing. Reading these disclosures at the time the instant invention was made, one of skill in the art may have reasonably inferred that the exact size of the spacer sequence chosen for any particular expression construct may vary, ranging anywhere from 5 to 1500 nts, without adversely affecting the outcome. Accordingly, it may be concluded that it would have been an obvious matter of design choice to make an expression construct with a spacer region of the size claimed in claim 12.

> MPEP 2144.04, subsection VI, C
> *In re Kuhle*, 526 F.2d 553, 188 USPQ
> 7 (CCPA 1975) (the particular placement of a contact in a conductivity measuring device was held to be an obvious matter of design choice). However, "The mere fact that a worker in the art could rearrange the parts of the reference device to meet the terms of the claims on appeal is not by itself sufficient to support a finding of obviousness. The prior art must provide a motivation or reason for the worker in the art, without the benefit of appellant's specification, to make the necessary changes in the reference device." Ex parte Chicago Rawhide Mfg. Co., 223 USPQ 351, 353 (Bd. Pat. App. & Inter. 1984).

In the instant case, one possible motivation to select a spacer sequence of the size recited in claim 12 may be to facilitate cloning. For example, Martinek et al., as part of their study of the use of inverted repeat expression constructs, state that 67 bp gaps were chosen to facilitate cloning (page 1719, 1st column).

Thus in the absence of evidence to the contrary, the invention as a whole would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made.

Response to Arguments

Again, Applicants arguments are directed to a claim no longer pending, claim 12. However, the arguments are addressed to the extent that they apply to current claim 45, now rejected over the art of record.

Applicants argue the references individually, stating that Driscoll et al. does not teach a spacer of 3–10 nucleotides and that the cited prior art does not describe any PLK-related construct and is mostly concerned with lower organisms. Thus, one skilled in the art would not know how to produce the claimed vectors for PLK-associated cancer therapy.

Applicants argue that Kennerdell and Martinek use a spacer that is 76 nucleotides long, which is less suitable for the claimed invention. Applicants assert that one would not be motivated to combine the cited prior art to arrive at the claimed invention.

Applicants cite a paper by Wakiyama et al. showing that spacers of 6 to 10 nucleotides are very effective.

Applicant's arguments filed 5/22/06 have been fully considered but they are not persuasive.

Contrary to Applicants assertion, and as pointed out in the previous Action, Kennerdell et al. do in fact teach an inverted repeat expression construct in which the sense and antisense strands are separated by a 5-base linker. This clearly falls within the claimed range of 3 to 10 nucleotides.

Taken together with the teachings of Driscoll et al., cited above, and Martinek et al. who teach a spacer range of from 300 to 1000 nucleotides and 67-nt, respectively, one of skill in the art may reasonably conclude that the length of the spacer may range as small as 5, 67, or 300

nucleotides in length.

While some spacers may provide for more efficient activity in some cases, one of skill in the art may reasonably expect that variations in the length and either the stem or the loop will produce changes in the relative activity of the hairpin expression vector.

Similarly, Elez et al. certainly provide a working example of an antisense oligonucleotide for gene-specific inhibition of human PLK1 in mice. Elez et al. provide ample motivation as well as an express suggestion to use their particular embodiment as well as others to silence PLK1 expression *in vivo* for the treatment of cancer.

Accordingly, the instant claim is rejected as obvious over the cited references.

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Claim 48 is rejected under 35 U.S.C. 103(a) as being unpatentable over Holtrich et al. (1994); Elez et al. (2000); and Driscoll et al. as applied to claims 32–35, 37, 39-44, 46, 49-51, 53-55, and 63 above, and further in view of Noonberg et al. (US Patent No. 5,624,803).

Claim 48 recites an RNA expression system according to claim 6, wherein the RNA promoter is U6 or H1.

Holtrich et al. (1994); Elez et al. (2000); and Driscoll et al. are relied upon for the reasons given above. These references do not teach a IR, hairpin expression construct comprising a U6 or H1 promoter.

Noonberg et al. disclose compositions and methods for generating U6 Pol III-driven expression cassettes for use in delivering antisense, ribozyme, and triplex formining

oligonucleotides intracellularly. It is taught that a distinct advantage of U6 Pol III promoters in such expression cassettes is the ability to generate oligos of a predetermined and well-defined length and sequence (column 12, lines 30-65). The Pol III U6 promoter is said to require only upstream sequences for initiation and is said to terminate cleanly upon reaching a string of 4-7 thymidine residues. Accordingly, the U6 Pol III promoter is well suited for generating high yields of short RNA oligos lacking extra, undesirable 5' sequences. The invention is summarized in column 13, lines 5-15. The invention is said to comprise a Pol III promoter such as the U6 promoter, a specific nucleotide sequence, which may designed so as to form a hairpin structure (columns 17-18), and a termination signal.

It would have been obvious to one of ordinary skill in the art to use U6 Pol III driven expression cassettes as taught by Noonberg et al. to generate short hairpin RNA expression vectors, as taught by Driscoll *et al.,* for inhibition of PLK1 gene expression in cells *in vitro* and *in vivo*.

One would have been motivated to create such compounds because Noonberg et al. expressly teach that U6 Pol III promoters are highly suitable for the purpose of expressing short, therapeutic RNA oligos intracellulary, as U6-type promoters terminate cleanly and result in oligos of a well defined sequence and length. In contrast, Pol II promoters are said to transcribe at lower frequencies, be cell-type specific, and generate transcripts with variable lengths and long polyadenylated tails (columns 17-18).

One would have a reasonable expectation of success given that Noonberg et al. fully describe the materials and methods necessary to generate U6-driven expression cassettes.

Thus in the absence of evidence to the contrary, the invention as a whole would have

been prima facie obvious to one of ordinary skill in the art at the time the invention was made.

Response to Arguments

Again, Applicants arguments are directed to a claim no longer pending, claim 15. However, the arguments are addressed to the extent that they apply to current claim 48, now rejected over the art of record.

In response to applicant's arguments against the references individually, one cannot show nonobviousness by attacking references individually where the rejections are based on combinations of references. See *In re Keller*, 642 F.2d 413, 208 USPQ 871 (CCPA 1981); *In re Merck & Co.*, 800 F.2d 1091, 231 USPQ 375 (Fed. Cir. 1986).

In the instant case, Applicants argue that Noonberg does not suggest or disclose a gene construct which can be used to reduce or inhibit the activity of PLK1.

However, the instant rejection is based on what the references as a whole would have taught or reasonably suggested to one of skill in the art at the time the instant invention was made.

While Noonberg alone does not teach all the limitations of the instant invention, Noonberg provides important guidance regarding the use of U1 and H1 promoters in general for driving the expression of antisense, ribozyme, and triplex forming oligonucleotides, which are art-recognized equivalents for purposes of gene expression inhibition. Noonberg specifically teaches that it is advantageous to incorporate self-complementary regions into the transcribed antisense RNAs to enhance their stability (col. 14). Accordingly, Noonberg teaches methods for generating not only antisense RNAs using U6 or H1-based expression constructs, but antisense

RNAs comprising hairpin loops.

Taken together with the teachings of Holtrich et al. (1994); Elez et al. (2000); and Driscoll et al., described above, one of skill would have been both well motivated and have had a reasonable expectation of success in making and using H1 or U6 driven expression constructs to inhibit PLK1 expression *in vivo* to inhibit tumor cell growth.

Accordingly, claim 48 is rejected as being obvious over the instantly cited references.

Response to Applicants' Arguments

Applicants' arguments presented on 5/22/06 not specifically addressed above are considered to be moot in view of Applicants' amendments to the claims and in view of the new and/or reiterated rejections stated herein, above.

Allowable Subject Matter

The prior art searched does not teach or suggest SEQ ID NO:30.

Conclusion

THIS ACTION IS MADE FINAL. Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after

the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the mailing date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Louis V. Wollenberger whose telephone number is 571-272-8144. The examiner can normally be reached on M-F, 8 am to 4:30 pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Peter Paras can be reached on (571)272-4517. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

Louis Wollenberger
Examiner, Art Unit 1635
September 15, 2006

SEAN McGARRY
PRIMARY EXAMINER
1635